

SELECTION FOR SPECIFIC HUMAN ANTIBODY BY ANTIGEN-INDUCED APOPTOSIS

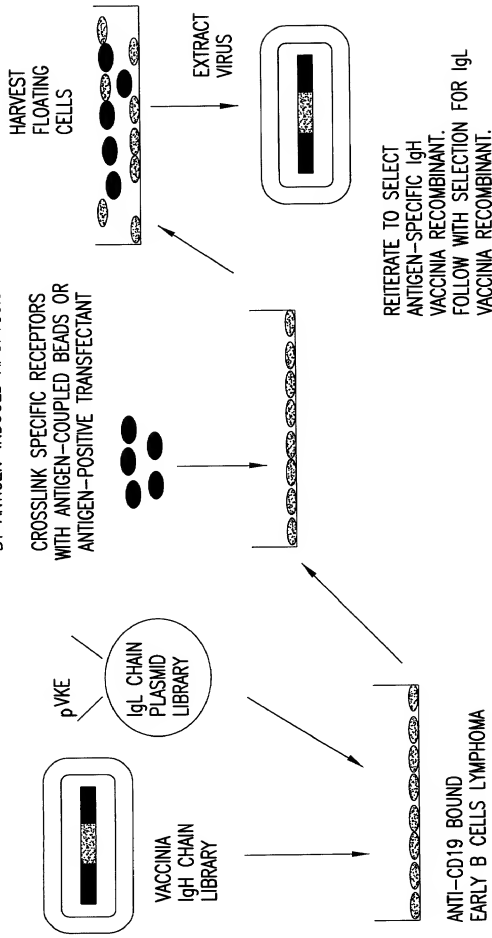


FIG.1

PREPARATION OF TARGET CELL LINES

EARLY B CELL PRECURSORS ARE TRANSFECTED WITH THE SELECTION CONSTRUCT

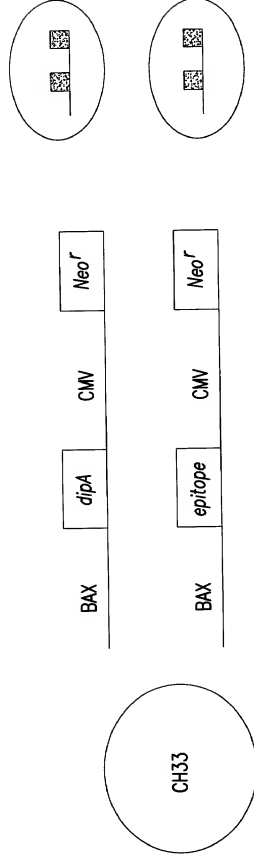


FIG.2A

VALIDATION OF MODIFIED CH33 TARGET CELL LINES

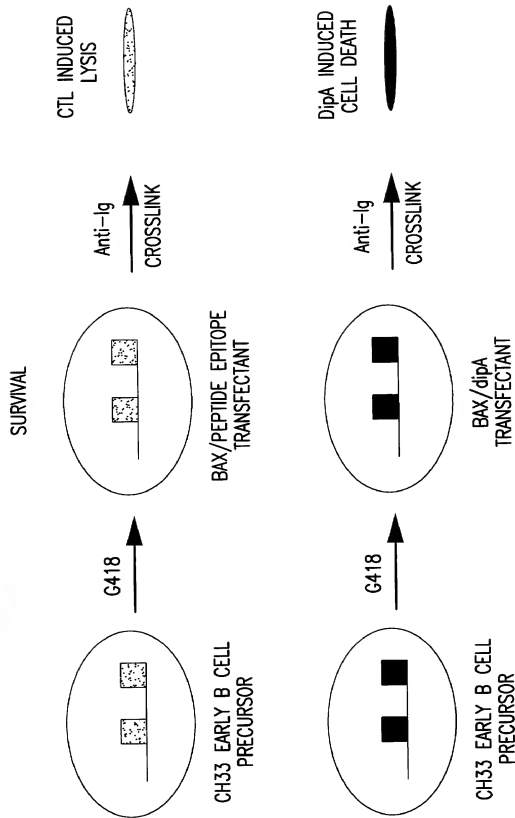


FIG.2B

CONSTRUCTION OF pVHE

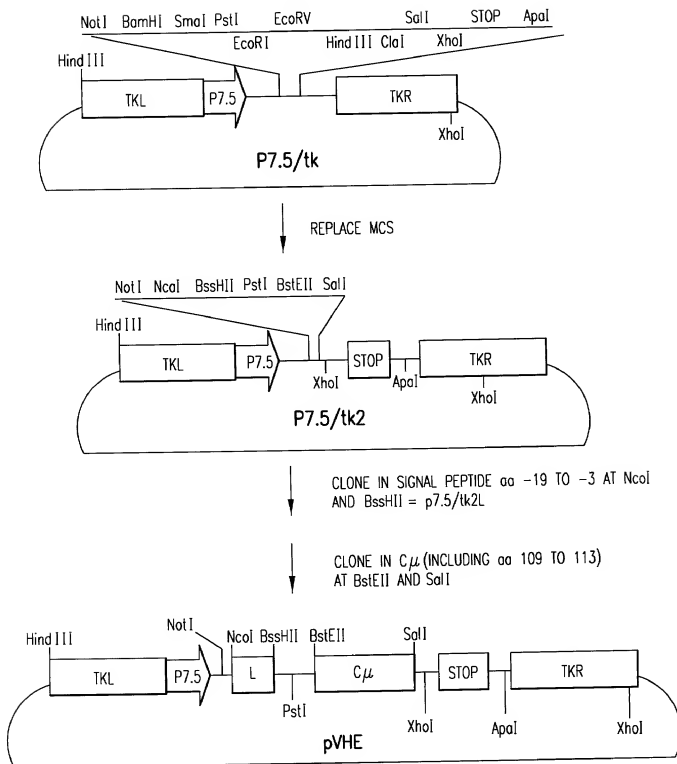


FIG.3

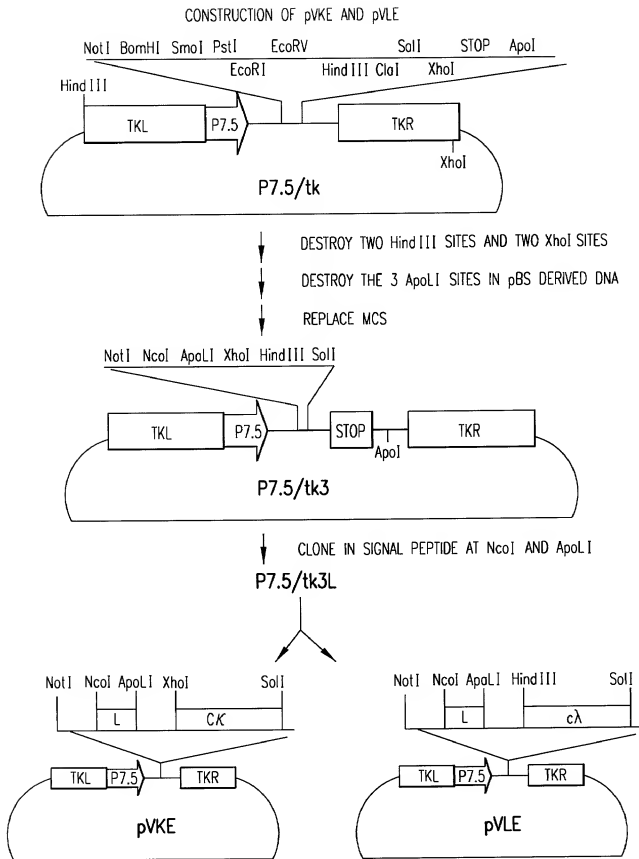


FIG.4

SELECTION FOR SPECIFIC HUMAN ANTIBODY BY ANTIGEN-DEPENDENT ADHERENCE

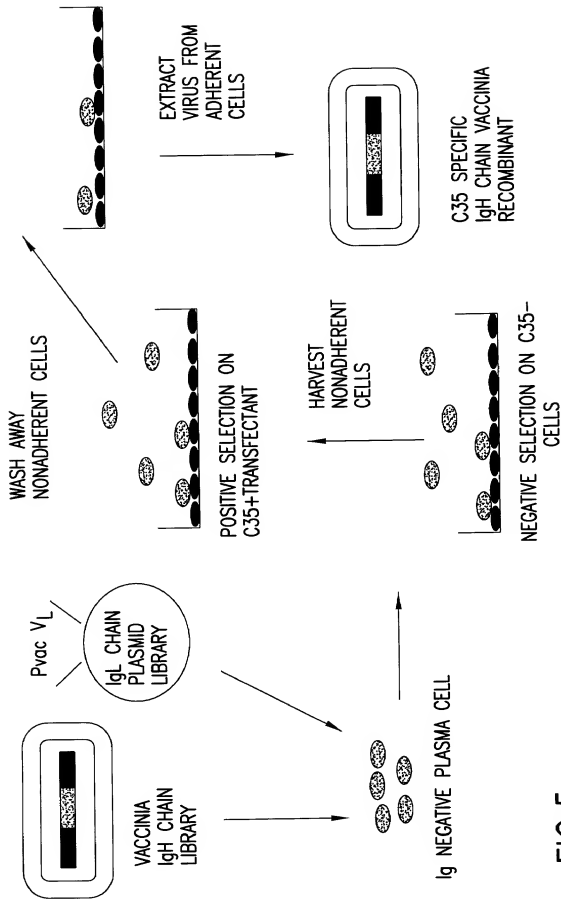


FIG.5

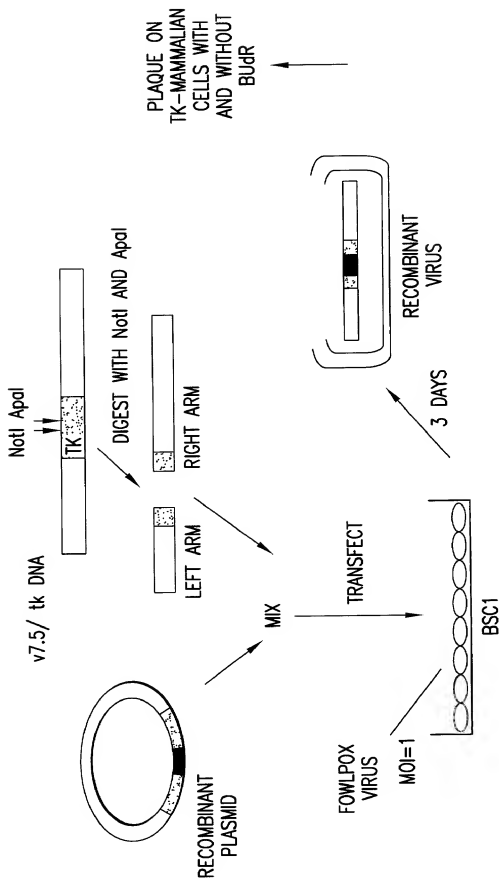


FIG. 6

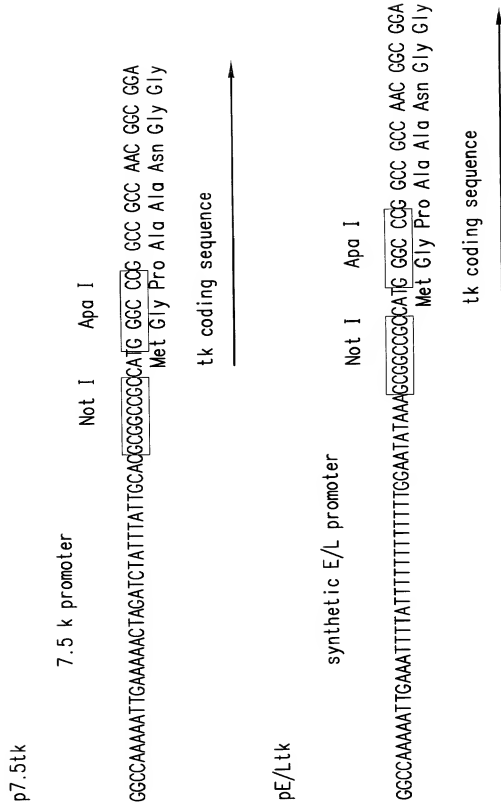


FIG.7

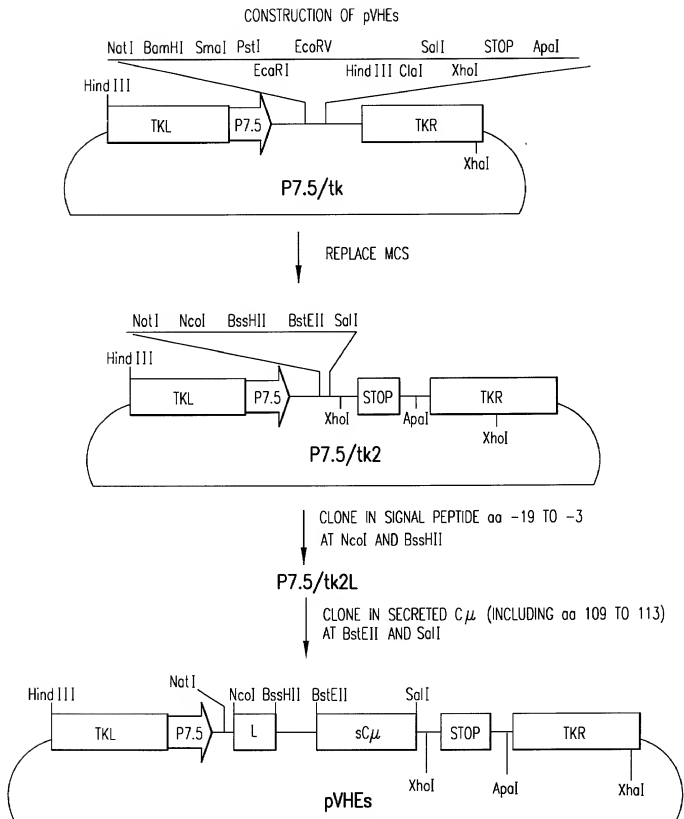


FIG.8

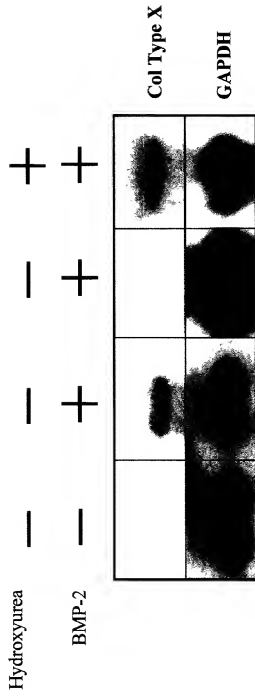
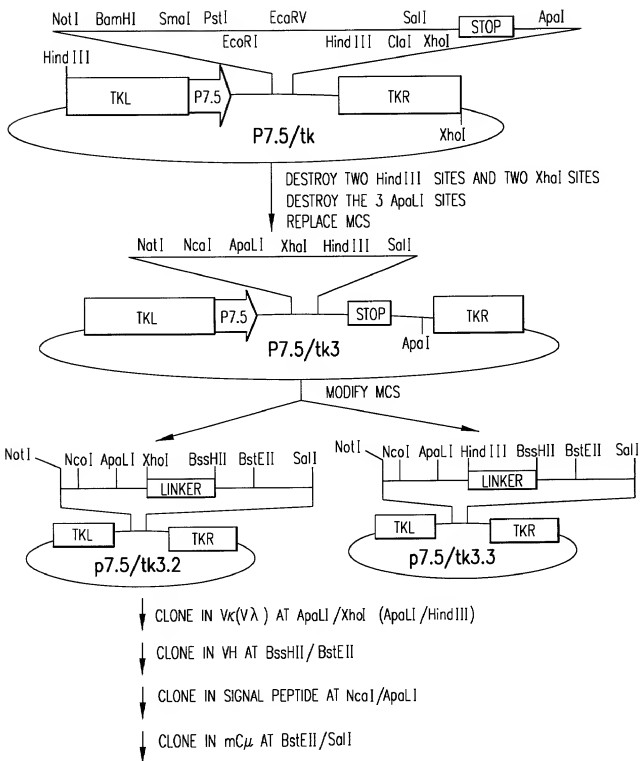
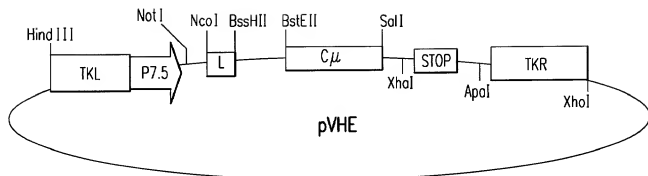


FIG.9



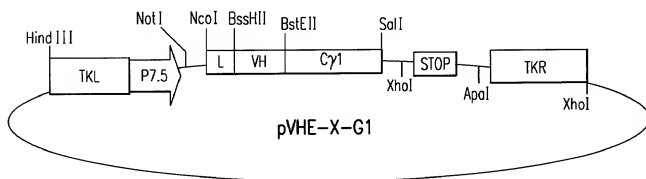
CONSTRUCTION OF scFV EXPRESSION VECTORS

FIG.10



↓ CLONE IN VH-X TO CONSTRUCT pVHE-X

↓ CLONE IN MODIFIED C γ 1



CONSTRUCTION OF pVHE-X-G1.

FIG.11

1. p7.5tk

7.5K PROMOTER NOTI APAI
5' - GGCCAAAATTGAAAACTAGATCTATTTATTCACGCGCGCCGCGCATGGGCCCGGCC -3'

2. p7.5/ATG0/tk

7.5K PROMOTER NOTI BAMI SMAI PSTI
5' - GGCCAAAATTGAAAACTAGATCTATTTATTCACGCGCGCGCGTGGATCCCCGGGCTGCAGGAA

SALI TRANSLATION TRANSCRIPTION
STOP CODONS STOP SIGNAL

TTTGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGGCCTAACCTAAATTTGTTTTGT

APAI

GGGCCCGGCC -3'

FIG.12A

3. p7.5/ATG1/TK

	7.5K PROMOTER	NOTI	START CODON	BAMHI	SMAI	PSTI
5' –	GGCCAAATTGAAAAC TAGATCTATTTATTGCACGGCGGCCATGGTGATCCCCCGGCTGCAGAA					
	SAL I		TRANSLATION STOP CODONS	TRANSCRIPTION STOP SIGNAL		
TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGGCCTAACTAACTAATTTGTTTTTGT						

APAI
GGGGCCGGCC –3'

FIG.12B

4. p7.5/ATG2/tk

7.5K PROMOTER NOTI START CODON BAMHI SMAI PSTI

5' – GGGCAAAAATTGAAAACTAGATCTATTTATTGCACGGCGGCCCATGAGTGGATCCCCGGGCTGCAGGAA

SALI TRANSLATION TRANSCRIPTION

STOP CODONS STOP SIGNAL

TTGGATATCAAGCTTATCGATACCGTGGACCTCGAGGGGGGCCCTAACTAACTAATTTTGT

APAI

GGGGCCGGCC –3'

FIG.12C

5. p7.5/ATG3/tk

	7.5K PROMOTER	NOTI	START CODON	BAMHI	SMAI	PSTI
5'–	GGCCAAAAATTGAAAAACTAGATCTATTTATTGCACGGCGCGCCATGACGTGGATCCCCCGGGCTGCAGGAA					

TRANSLATION	TRANSCRIPTION
STOP CODONS	STOP SIGNAL

TTCGATATCAAGCTTATCGATACCGTCGACCTCGAGGGGGCCTAACATAATTTGTTTTTG

APAI

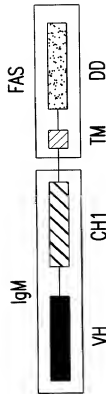
GGGGCCGGCC –3'

FIG.12D

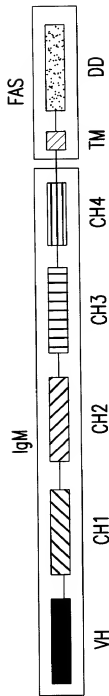
CONSTRUCTION OF IgM-Fas FUSION PRODUCTS

CONSTRUCTS:

(a) VH-CH1-TM-DD



(b) VH-CH1-CH2-CH3-CH4-TM-DD



(c) VH-CH1-CH2-CH3-CH4(TM)-DD

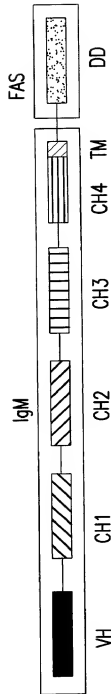
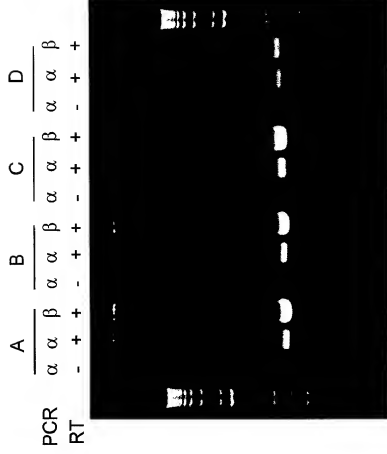


FIG.13



Expression of Ig α and Ig β in the transfected COS7 and HeLaS3 cell lines.

FIG.14